## JAN 10 1917

## AN ICHTHYOBELLID PARASITIC ON SAND WHITING.

## On an Ichthyobdellid parasitic on the Australian Sand Whiting (Sillago ciliata).

By

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With Plates 1 and 2, and 6 Text-figures.

#### INTRODUCTION.

IN August, 1912, during a visit to the Fish Hatcheries Institution, Port Hacking, an inlet south of Port Jackson, New South Wa'es, my attention was called to a parasite which, I was informed, had for some time past caused the death of sand whiting kept in a large spawning pond. An examination of this parasite showed it to be a marine leech.

The history of the infestation, so far as I have been able to ascertain, is as follows.

It has been the custom for some years past to stock a large sea-water pond of the Institution with two or three dozen sand whiting taken from the shoals in the vicinity. This appears to have been done several times, and on each occasion the fish were killed by the attacks of these leeches.

The fish, shortly after being placed in the pond, sickened, developed large ulcerated patches on their integument and died. Within ten weeks of being introduced into the pond most of the fish would be in a moribund condition.

It was easy to know when the fish were seriously affected, for then they would swim very close to the surface, and on their sides. An examination of a badly infested fish usually

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showed about a hundred leeches in various stages of development, ranging in length from 1 to 13 mm.

The parasites were found on the fins—pectoral, pelvic, dorsal, and caudal; their presence was also noticed around ulcerated patches on the sides of the fish, and a few were found in the proximity of, but not on, the gills.

Owing to their transparent nature, and the mucous secretions of the fish, they were not so readily visible as their highly pigmented form seen when magnified would suggest.

On the occasion of the first visit in August I secured several hundred specimens, and in March of the following year, having determined to work out the anatomy and systematic position of this leech, I again visited the Fish Hatcheries, but found that all the sand whiting had died. Dredging giving no results, arrangements were made to again place a number of the fish in the pond in order to obtain living specimens of the leech.

Owing to an oversight, I was not informed of the state of these fish until all save one had died. This remaining fish was caught on June 8th, 1913, and was found but slightly infested. Most of the leeches obtained died in the first few days, but two hardy specimens were kept alive for three weeks, and these served for an extended intra vitam examination, for taking photographs, and for making a coloured drawing.

Owing to the transparent nature and small size of the leeches, the details of the Blood-vascular, Nephridial, and Cœlomic systems could be followed, and this with only the slight compression of the leech produced by the pressure of a thin cover slip.

A number of photographs of the living form were secured showing these systems in detail, and have been of value in this work. The pond in which these fish were kept under conditions which so favoured the increase of the leeches was a large one, being about  $50 \times 100$  ft. The water in it varied in depth from 2 to 6 ft., and was changed by means of values, the water being run off at low and replaced at high tide.

About a dozen species of fish were kept in it: as well as the sand whiting (Sillago ciliata), there were present Pagrosomus auratus, Chrysophrys australis, and Caranx georgianus.

In no case were these leeches found on any fish other than the sand whiting. That this pond formed a favourable place for the development and increase of parasites was also shown by the fact that most of the fish, except the sand whiting, were infested by ectoparasitic Trematodes; these will in due course be described by Dr. S. J. Johnston, of the Sydney University.

At the beginning of February, 1914, I had the opportunity of examining many hundreds of sand whiting netted by fishermen or caught by line in the estuarine and ocean waters of Wreck Bay, about 100 miles south of Port Jackson. Among these fish it was rare to find an individual which did not have from two or three to half a dozen specimens of this leech. I examined a large number of other species of fish netted along with the sand whiting, but never found them infested with this or any other leech.

It was found necessary to create a new genus to contain this leech, which will be described under the name of Austrobdella translucens.

#### Austrobdella gen. nov.

Definition.—Small marine leeches with well-defined neck and body regions. The body cylindrical in the young, but much flattened in the adult. The lateral parts of the body below the clitellum bulging out and forming a shoulder-like appearance. Somite of six annuli. No pulsating vesicles present, their place being taken by a continuous contractile lacuna placed on either side outside the body musculature. Dorsal and ventral median lacunæ present, communicating by segmental lacunæ. Three pairs of pouches present in tho thick-walled intestine, a fourth pair being represented by a flexure of the gut. Testes five pairs. Eyes one pair.

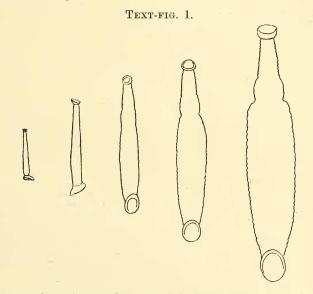
Type species A. translucens, mihi. Type specimen in the Australian Museum, Sydney, No. W. 403.

## EXTERNAL FORM.

The material on which this work is based consists of a large number of specimens ranging from the young sexually immature to the adult form. From a number of specimens it has been possible to arrange a complete series of individuals to show the change of form during growth. I would lay stress on this very marked change in form, and my observations have convinced me of the likelihood of the younger forms of similar marine leeches being described as new genera or species in the absence of a series of individuals linking up the young to the adult.

In Text-fig. 1 are shown the outlines of specimens which measure 1.5, 3, 4, 5.5, and 7.5 mm. long, to illustrate the change in form which takes place during the growth of this leech. Young forms possess a cylindrical body, and the chief change in shape during growth is due to the great lateral development of the body posterior to the clitellum. The appearance of a specimen 1.5 mm. long is shown in Pl. 1, fig. 3. When this leech is 4 mm. long the testes become mature. At this stage the body is cylindrical (Pl. 1, fig. 4), there is but a faint indication of the clitellum, and the ovaries are quite undeveloped. In 6 mm. specimens maturation divisions are seen in the ova. At this stage the clitellum has become enlarged and is now well marked.

The young of Austrobdella have the anterior sucker of the same diameter as the cylindrical body and the posterior sucker nearly twice that size (Pl. 1, fig. 3). When a length of 4 mm. is reached the anterior and posterior suckers approach one another more closely as regards size and have a diameter a little less than the greatest diameter of the body (Pl. 1, fig. 4). When the leech has a length of 5.5 mm. (Text-fig. 1) the typical form of the adult is foreshadowed. At this stage the clitellum becomes evident, and owing to the increased lateral development that part of the body posterior and lateral to it presents a shouldered appearance; but the body is only slightly flattened. When a length of 7.5 mm. (Text-fig. 1) is reached the lateral development is quite pronounced, causing a more flattened form, and the suckers have assumed the ratio shown in the figure of an adult specimen, 9 mm. (Pl. 1, fig. 5).



Outlines of specimens of Austrobdella translucens which measure 1.5, 3, 4, 5.5 and 7.5 mm. long. The figure shows the change in shape during the growth of this marine leech.

I give below a table to show the measurements at different stages of development :

|   | Length. | Breadth below<br>Clitellum. | Height. | Diameter<br>Oral Sucker,               | Diameter<br>Posterior Sucker. |
|---|---------|-----------------------------|---------|--|-------------------------------|
| ( | 1.5 mm. | ·13 mm,                     | ·13 mm. | ·13 mm.                                | -20 mm.                       |
|   | 4.0 mm. | ·52 mm.                     | ·5 mm.  | ·30 mm.                                | ·41 mm.                       |
|   | 7.5 mm. | 1.35 mm.                    | ·7 mm.  | ·55 mm.                                | ·91 mm.                       |
|   | 9.0 mm. | 3.25  mm.                   | 1.0 mm. | $\cdot 8 \times \cdot 5 \text{ mm.} =$ | $1.75 \times 1.1 = 1$         |
| G | 100     | - COLE                      |         | $\cdot 65 \text{ mm}.$                 | 1.42 mm.                      |

The measurements given for the 9 mm. stage are from the specimen which is shown in Pl. 1, fig. 5. The following additional measurements are also from this specimen:

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All these measurements are taken from leeches killed by means of boiling corrosive acetic solution while in a quiescent condition.

In describing Platybdella michaelseni, Johansson (1911) states that the larger of two specimens secured was 6.9 mm. long. He found in this specimen that the testes were ripe but the ovaries slightly developed, despite the presence of spermatozoa. He therefore considers that this species would not attain a much greater size. However, if my observations on the growth of Austrobdella are found to be generally applicable to marine leeches, the species he describes might grow to nearly twice the length given.

#### COLORATION.

The transparent nature of the body of Austrobdella allows the beautiful pigmentation to be clearly seen. The drawing (Pl. 1, fig. 1) is a careful representation in black and white of a living specimen, viewed from the ventral surface, as seen by transmitted light.

The specimen depicted is a young extended form in which neither the clitellar region nor the lateral parts of the main body region are as well developed as usual. This individual was made use of owing to its having retained its colour in captivity better than its fellows.

On the ventral surface most of the pigment cells are seen to be wrapped round what appears to be a tube. These cells, which are reddish-brown in colour, are in the walls of the ventral lacuna; their stellate nature is well represented. Similar pigment cells are seen in relation to the ejaculatory canals; these are here, as in most specimens, rendered conspicuous by their dark colour.

As well as the reddish-brown pigment cells mentioned, a smaller number of cells of varying shades of purple are present; some of these show through from the dorsal surface. Large and more diffuse yellow cells are scattered about the surface, and at the posterior region of the body certain of the cocoon glands have a light brown colour.

Viewed from the dorsal surface the lighter coloured pigment cells of the neck region are seen to be placed in a single lateral and a medial double row.

The darker cells are found in a well-marked single row along the mediad wall of the contractile lacuna and are plentiful, but without any apparent regular arrangement in the dorsal body wall.

The lighter cells appear as five irregular rows in each side of the body.

In a living specimen the two eye-spots are very conspicuous. They are of a rich dark reddish brown colour, larger than any of the pigment cells and characterised by their regular outline in place of the fringed appearance of the pigment cells. In the drawing they are shown as seen from the ventral surface through the body tissues.

A microscopic examination of the caudal fin of a living sand whiting to which a leech was attached showed a remarkable similarity in colour and arrangement of the pigment cells of the two animals.

So far as my observations go the pigment cells lose their fringed character in strong light and become more regular in outline.

#### MOVEMENTS.

During my observations of living specimens I never saw an individual swimming, neither could I, by dropping the leeches into salt water, cause any swimming motion. They would fall straight to the bottom of the beaker and then move slowly along the surface of the glass by leech-like movements. These observations were made on leeches ranging in size from 2 to 12 mm. They have a bearing on the question of the deposition of the cocoon and the manner of infestation of sand whiting by young leeches. Johannson (1898), writing

about A branchus (a genus allied to Austrobdella), correlates the fact that the members of this genus are unable to swim, with the fact that they infest fish which live in shallow water among algal growths and so afford an opportunity for leeches to creep on to them. He also supposes that when the time comes for depositing the cocoon Abranchus drops to the bottom and fixes its cocoon in seaweed. I have not ascertained the method of depositing the cocoon in Austrobdella, but would point out that the fish on which it lives gets a great deal of its food by burrowing in the sand.

#### ANNULATION.

A typical somite consists of three annuli, and in the adult these three annuli are divided so that the somite is of six annuli. In young forms the somite consists of the three primitive annuli; this is followed by a stage in which there are five annuli to the somite, due to the division of the annuli anterior and posterior to the annulus in which the nerve ganglion is situated. This stage of five annuli to the somite may persist in specimens which are quite large, but in the largest specimens all the somites in the testicular region have six annuli. Thus it happens that specimens 6 mm. in length are seen, in which the primitive annulus containing the nerve gauglion is still undivided. It is owing also to this subsequent division of primitive annuli that a difficuly arises in regard to the number of annuli to be seen in the neck region, for these increase in number with the length of the specimen. In a leech of 4 mm, the male pore is between the 15th and 16th primitive annuli and the female pore between the 17th and 18th annuli. At this stage the somite in the testicular region is still trimerous. In a leech of about 9 mm. there are found twenty-three or twenty-four annuli in front of the male pore. The primitive annuli which divide to form the new annuli are the 4th, 5th, 7th, 10th, 11th, 13th, 14th, and 15th (Text-fig. 2). In regard to the total number of annuli, my determinations have been varied owing to the fact that

the annulation of the posterior part of the leech is very indistinct, so that in specimens in which the annulation of the neck

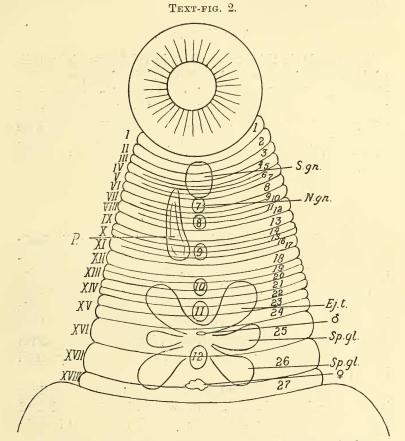


Diagram showing the character of the annulation of the preclitellar and clitellar regions of Austrobdella translucens and the relations of the nerve ganglia, proboscis, and sexual organs. S. gn. Subæsophageal nerve ganglion. N. gn. Nerve ganglion. P. Proboscis. Ej.t. Terminal part of ejaculatory canal. Sp. gl. Spermatophore glands. A Male opening.  $\heartsuit$  Female opening. Roman numerals indicate primitive annuli, ordinary numerals the subdivision of the primitive annuli.

and mid-body can be seen, that of the posterior part of the body is not evident.

The relations of the nerve ganglia, male and female openings, and the male glands to the neck annuli are shown in Text-fig. 2.

#### THE EPIDERMIS.

The cuticle is thin,  $2 \mu$  or less in thickness. The cells of the epidermis are irregularly cubical in shape, measuring about 8  $\mu$ . Towards the anterior and posterior suckers they become more cylindrical and have a length of  $14 \mu$  by  $5 \mu$ wide. On the ventral surface towards the median plane these epidermal cells are not so numerous. But a few of them are slightly enlarged and converted into gland cells with ducts opening on the surface.

## Hypodermal Glands.

There is developed on the lateral margin of the main body region, partly surrounding the contractile lacuna, a remarkable layer of cells, three or four deep and of great size (Pl. 2, fig. 9, *L. gl.*). Each is a unicellular gland and its duct opens on the ventral surface. The largest of these cells has a diameter of 63  $\mu$  and the size ranges from this to about 15  $\mu$ . The shape varies considerably, but in all the secretion space passes gradually into the duct, which has a very small lumen.

The nucleus varies a good deal in appearance; generally it is more or less spherical, but frequently elongated and twisted; it has many chromatin particles staining heavily with hæmatoxylin.

These glands appear to correspond to certain cells, not so well developed, called the lateral glands in Branchellion by Sukatschoff (1912).

As is general in Ichthyobdellids, both mucous and albuminous unicellular glands are present in the two suckers.

In the oral sucker the albuminous glands are arranged around the dorso-lateral three-fourths of the sucker. They are placed inside the circular and longitudinal musculature of

the sucker wall, and, measuring about 20  $\mu$  in diameter, are about half the size of the same glands in the posterior sucker. Among them are placed the smaller mucous glands, and both types of glands open by ducts on the concave surface of the sucker. In the posterior sucker these glands are distributed over the whole sucker. The mucous glands are found also forming a pharyngeal group with ducts opening into the pharynx, and they extend posteriorly and lie among the salivary glands but nearer the body wall; they are not present below the glandular œsophageal pouches.

### CLITELLAR GLANDS.

As has been remarked by various authors, the development of the unicellular glands in the Ichthyobdellids is most remarkable. To quote Bourne (1884): "They attain relatively, and in Pontobdella and Branchellion actually, huge dimensions."

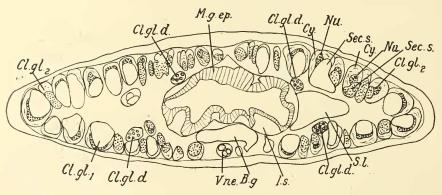
In this leech the largest among these cells have a diameter of 170  $\mu$ , and, if they are situated far back in the body, a duct 7 or 8 mm. long. The clitellar glands extend in the body region from the posterior sucker to the beginning of the clitellum.

They are placed inside the body wall musculature and occupy practically all the space between it and the alimentary, reproductive, and lacuna systems. Text-fig. 3, drawn from a transverse section through the region of the thick-walled middle gut, shows the degree of development of these glands (Cl. gl. 1, Cl. gl. 2).

Their ducts open all round the clitellum, that is from the level of the 10th nerve ganglion to midway between the 12th-13th nerve ganglia. Sukatschoff's (1912) excellent work on these glands showed that in Branch ellion there were three types producing different secretions, and he gives a description of extraordinary development of the branched nucleus, which may come to measure 336  $\mu$  and which he compares (p. 488) with other examples of nuclei of this type in the animal kingdom.

In Austrobdella I shall content myself for the present with the appearances presented upon staining with Ehrlich's hæmatoxylin and eosin. By this means there are clearly differentiated two types of clitellar glands. The larger of these cells (Text-fig. 3, *Cl. gl.* 1), which are bowl-shaped or more elongated, are characterised by their large secretion space filled with a homogeneous substance showing a finely granular nature when highly magnified, and staining a light pink with eosin. These cells, which have an average diameter

## TEXT-FIG. 3.

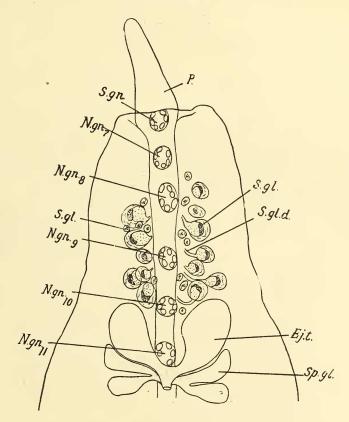


A transverse section through the thick-walled, middle-gut region of Austrobdella translucens ( $\times$  40), showing the arrangement, development and character of the clitellar glands. The relation of the intestinal sinus to the epithelium of the thickwalled middle gut is shown. A portion of a segmental lacuna of one side is present. The blind gut has been cut at its narrowest part. Cl. gl. 1. Clitellar gland of the first type. Cy. Cytoplasm. Nu. Nucleus. Sec. s. Secretion space. Cl. gl. 2. Clitellar gland of the second type. Cl. gl. d. Groups of ducts of clitellar glands. V. ne. Ventral nerve cord. B. g. Blind gut. S. l. Segmental lacuna. I. s. Intestinal sinus. M. g. ep. Epithelium of thick-walled part of middle gut.

of  $110 \mu$ , have the bulk of their cytoplasm at the peripheral end in a layer, which is 30 or 40  $\mu$  thick, and which lines, as it were, the secretion space, becoming finer towards the ducts. This cytoplasm is of very coarse structure, and the nucleus, which is rich in large chromatin particles, has an irregular shape, as described by Sukatschoff in Branchellion, and is placed next to the secretion space (Text-fig. 3, Cy., nu., sec. s.).

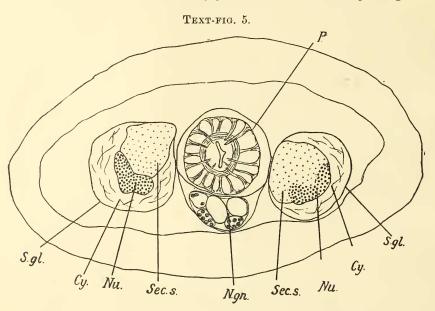
The other type of clitellar gland cell (Text-fig. 3, Cl. gl., 2) is distinguished by the intense staining of the secretion by

TEXT-FIG. 4.



Drawn from a whole mount of a specimen of Austrobdella translucens 6 mm. long ( $\times$  100) which was killed when the proboscis was protruding. The figure shows the character of the salivary glands and their relations to the nerve ganglia. *P.* Proboscis. *S. gn.* Subæsophageal nerve ganglion. *N. gn.* 7. Seventh nerve ganglion. *S. gl.* Unicellular salivary gland. *S. gl. d.* Unicellular salivary gland duct. *Ej. t.* Terminal part of the ejaculatory canal. *Sp. gl.* Unicellular spermatophore glands.

eosin. This secretion is coarse and consists of particles of  $2 \mu$  in diameter. These cells, which are a good deal smaller than those of the first type, have a diameter of about  $60 \mu$ , and are placed in between the larger cells beneath the body wall musculature, as well as internal to the larger cells, and a few are found outside the salivary glands with their ducts passing



Transverse section through the preclitellar region of a specimen of Austrobdella translucens 7 mm. long ( $\times$  186). The drawing shows the character of two of the unicellular salivary glands and their relatively large size. The position and character of the nucleus which is next to the secretion space is also shown. S. gl. Unicellular salivary gland. Cy. Cytoplasm. Nu. Nucleus. Sec. s. Secretion space. N. gn. A ganglion of ventral nerve cord. P. Proboscis.

back to open on the clitellum. The nucleus, which is irregular in shape, is either next to the secretion space or in it, and separated from the cytoplasm. The ducts of these two types of cells, filled with their characteristic secretion, are gathered into four bundles, which, just before the clitellum, are placed two in the dorsal and two in the ventral region.

The ducts of the larger glands have a diameter of  $8 \mu$ , the smaller  $6 \mu$ .

## SALIVARY GLANDS.

The salivary glands, as shown in Text-fig. 4 (S. gl.) are placed between the upper level of the eighth, and the lower level of the tenth nerve ganglion. On either side of the proboscis there are about five or six giant-cells and a number of smaller ones. The large cells have a diameter of about  $120 \mu$ , and other cells range in size from that down to  $20 \mu$ . Their ducts curve at right angles to the body of the cell and enter the base of the proboscis. The character of these cells and their relatively huge size is shown in Text-fig. 5. The position of the nucleus, which is next to the secretion space, is also shown.

## THE CŒLOMIC SYSTEM.

We owe to Johansson (1898) and to Selensky (1906) our main knowledge of the cœlomic system of the Ichthyobdellids other than Pontobdella, Branchellion, and Ozobranchus. The former in 1896 pointed out the great value of the knowledge of this system in systematic work, giving a number of examples of the chief features in different Ichthyobdellids. This leech, while possessing the main features of the cœlomic system as described for Piscicola and Callobdella, diverges widely in certain respects. This divergence is most marked in that part which corresponds to the contractile vesicles of Piscicola and Callobdella.

In Austrobdella, in place of a lateral row of such vesicles, there is a continuous contractile lacuna. This lacuna occupies the position of the contractile vesicles, as described for allied genera, lying laterally just beneath the skin outside the muscle layer. On either side it extends from the level of the proboscis to the level of the anus, but is contractile only in the region of the testes and the thick-walled intestine. This genus differs also in wanting altogether the lateral

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lacuna found in a number of Ichthyobdellid leeches; a fact of considerable importance when considered in relation to the direction of the flow of lymph in the body-cavity. Proceeding to a detailed examination there are found the following lacunæ: Dorsal, Ventral, Contractile, and the Segmental Lacunæ.

I shall follow Oka and Selensky in the use of the term "lacuna" in place of "sinus" in dealing with the various portions of the cœlome, reserving the term "sinus" for the blood-space found round the intestine called by Johansson the "intestinal lacuna."

The Dorsal Lacuna may be considered in two parts, the testicular and the intestinal portions. It extends from the beginning of the testes to the anal region. In the testicular region it contains the dorsal blood-vessel (Pl. 2, fig. 8, *Dl.*); in the intestinal region it surrounds the thick-walled intestine and the blood-stream in relation with it.

Posteriorly the intestinal portion communicates with the ventral lacuna in the anal region. In each segment the dorsal lacuna communicates by a pair of segmental lacunæ with similar extensions of the ventral lacuna (Pl. 2, fig. 8, S. l.). The wall of the testicular portion of the dorsal lacuna has a thin well-defined membrane with small elongated nuclei, which are scarce. Beneath this membrane are a few muscle-fibres.

The musculature is increased where the dorsal blood-vessel is fused with the wall of the dorsal lacuna.

Frequently the dorsal lacuna is divided into two parts by the formation of septa placed dorsally and ventrally to the dorsal blood-vessel (Pl. 2, fig. 10, *Sep. d. Sep. v.*). Such septa generally begin with the origin of the valves of the dorsal vessel, but do not extend to the preceding or succeeding valve-origin; so that at certain places the dorsal blood-vessel lies free in the dorsal lacuna.

The Ventral Lacuna (Pl. 2, fig. 8, v.l.) occurs as a tube, which varies in size and runs from the anal region to the proboscis, ventral to the alimentary canal. Throughout

its extension it contains the ventral nerve cord and the ventral blood-vesssl.

Anteriorly it is considerably expanded and entirely surrounds the proboscis and related organs.

A considerable expansion of the lacuna also contains the ovaries, and again it is dilated in the region of the posterior ganglionic mass. A pair of segmental communications is given off at the level of each nerve ganglion, joining with those given off more posteriorly in each segment by the dorsal lacuna.

Everywhere the ventral lacuna is lined by a membrane of the same character as that of the dorsal lacuna, but the muscle-fibres are few.

The Segmental Lacunæ, as already mentioned, extend from the dorsal and ventral lacunæ. They unite towards the lateral region of the body, just past the testes of each segment, and are also found in the thick-walled intestinal region. Reference to the figure of the Cœlomic System (Pl. 2, fig. 8) will aid the explanation of the course and branching of the segmental lacunæ.

Immediately after the junction of the dorsal and ventral extension, the lacuna divides and more laterally each division again divides in two; of the four ultimate branches, the two inside ones unite and open into the contractile lacuna (Pl. 2, figs. 8, 9, S. l.).

The two outside branches unite with the outside branches of the preceding and succeeding segmental lacunæ respectively. Thus it follows that in each segment there are two openings of the segmental lacunæ into the contractile lacuna. These openings into the contractile lacuna are furnished with musclefibres of annular arrangement and sphincter action.

The lining of the segmental lacunæ is a continuation of that of the dorsal and ventral lacunæ; no muscle fibres are found in their walls.

Contractile Lacunæ.—These extend on either side from the level of the base of the proboscis to the level of the anus, but are only contractile from the beginning of the testicular region. As already stated, their chief feature is their

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extension as tubes in place of the row of vesicles found in allied genera. They possess, as shown in Pl. 2, fig. 9 (C. l), the character of a series of pouches; their walls are furnished with delicate muscle fibres. Anteriorly the contractile lacunæ cease to be contractile at the level of the neck but are continued forward as non-contractile parts to the anterior sucker. I have not determined their course past this point, but they certainly do not appear to break up into capillaries.

Posteriorly their relations are more important. At the level of the posterior sucker they curve sharply, and, passing ventral to the two branches of the dorsal vessel, open into the lacuna in the anal region. They receive on either side of each segment the two openings of the segmental lacunæ (Pl. 2, fig. 8, C. l.). Dorsally in each segment they give off three or four pairs of capillaries (Pl. 2, fig 8, Cap. l.); these run parallel to one another, just outside the muscle layer. In one living specimen I observed these opening into the dorsal lacuna, but was unable to demonstrate this in other specimens.

In Callobdella Johansson has seen three or four pairs of capillaries in each segment going from the dorsal lacuna and stretching into the surrounding tissue.

## The Circulation of the Lymph.

In the description of the circulation of blood in the vessels mention will be made of the contraction of the pouches of the thick-walled intestinal region. By these contractions a space is produced between the wall of the intestinal sinus and the wall of the intestinal lacuna, which is immediately filled by the lymph flowing in from the lacuna formed in the anal region by the fusion of the dorsal and ventral lacunæ. When the intestinal sinus is again in the condition of diastole, the lymph is seen to be forced out of the dorsal lacuna and to flow into the segmental lacunæ. At the spot where the dorsal and ventral extensions of the segmental lacunæ join a great deal of regurgitation takes place. The lymph corpuscles

are seen to be violently hurried in various directions. Some are driven into the extensions of the segmental lacunæ on either side, some towards the ventral lacuna, but the majority pass through the openings leading to the contractile lacuna. These are immediately closed by the annular muscle fibres (Pl. 2, fig. 9, S. m. f.), which function as a sphincter when the contractile lacuna begins to contract. This contraction is from before backward and the lymph is carried to the lacuna, lying in the anal region, formed by the fusion of the dorsal and ventral lacunæ, whence it flows again into the dorsal lacuna. The contraction of the contractile lacuna immediately follows the diastole of the intestinal sinus, and the dorsal blood-vessel in the testicular region; so that it is seen, in a leech freshly taken from an ocean whiting, to take place about thirty times per minute.

In this description of the cycle of the circulation of the lymph, it will be observed that the events taking place in the ventral lacuna are not mentioned. This is due to the relative stagnation of the lymph in this lacuna. Despite repeated attempts to find a definite direction of flow in the ventral lacuna, I have been unable to observe anything more than a great deal of regurgition both in this and the ventral parts of the segmental lacunæ. Frequently strong currents carry lymph corpuscles from the dorsal segmental lacunæ far into these lacunæ, and these, and other corpuscles therein, are kept in constant movement by eddies. But there appears to be no such definite direction of flow as is present in the dorsal and contractile lacunæ.

# Comparison with the Lymph Circulation of other Leeches.

Salensky's (1906) description of the valve arrangement in the side vesicles of Piscicola makes evident the course of the circulating lymph in this leech and affords an interesting comparison with the circulation above described.

In Austrobdella I consider the contractile lacunæ, on account of their subcuticular position and pouched character,

to be homologous with the paired vesicles of other Ichthyobdellid leeches. They perform the functions of the side vesicles and lateral lacunæ of Piscicola-like forms. In place of the valve described by Salensky, there are found openings guarded by sphincter-muscle fibres.

The contraction of the dorsal vessel in the testicular region is not caused by the lymph flowing back from the lateral regions, as described by Johansson (1896 b) for Callobdella, for here in the segmental lacunæ the flow is always to the contractile lacunæ. It is mainly caused by the flow of lymph from behind and partly by the contractility possessed by the blood-vessel itself.

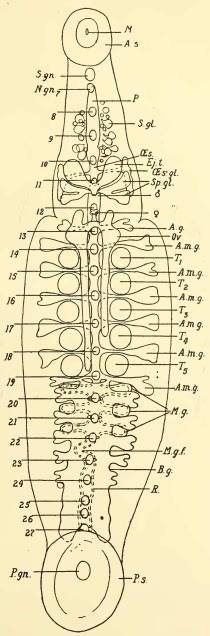
## ALIMENTARY SYSTEM.

Here I shall employ the terms used by Sukatschoff (1912) in his excellent description of this system in Branchellion. The mouth opening is placed slightly towards the dorsal side of the anterior sucker and leads into the short pharynx (Textfig. 6, M.). Following the pharynx is the œsophagus, which contains the proboscis characteristic of the Rhychobdellid leeches and capable of being protruded by the eversion of the pharynx and œsophagus. There follows then the entodermal anterior gut.

The proboscis, when retracted, occupies somites 7, 8, and 9 (Text-figs. 2 and 6, P.). The proboscis sheath consists of a thin ectodermal epithelium covering a number of longitudinal nuscle fibres. It corresponds very closely to the same structure described in Branchellion by Sukatschoff—the

<sup>TEXT-FIG. 6. — Diagram of the Nervous, Alimentary and</sup> Reproductive Systems of Austrobdella translucens.
(× 20), A.g. Cæca of anterior entodermal gut. A.m.g. Cæca of anterior thin-walled part of middle gut. A.s. Anterior sucker. B.g. Blind gut. Ej.t. Terminal part of ejaculatory canal. M. Mouth opening. M.g. Cæca of thick-walled part of middle gut. M.g.f. Flexure of thick-walled part of middle gut, representing a rudimentary pair of cæca. N.gn.7. Nerve ganglion 7, etc. C.s. Cesophagus. CEs.gl. Cesophageal glands. Ov. Ovary: P. Proboscis. P.gn. Posterior ganglion of ventral nerve cord. P.s. Posterior sucker. R. Rectum. S.gl. Salivary glands. Sp.gl. Spermatophore glands. S.gn. Subœsophageal ganglionic mass. T. Testis.

TEXT-FIG. 6.



only difference being that the nuclei of the ectodermal epithelium cells are more numerous, there being always one and sometimes two between each bundle of muscle fibrils (compare Sukatschoff, fig. 73).

The proboscis is covered by a thin epithelium, beneath which are the longitudinal muscle fibres arranged in similar fashion to those of other Ichthyobdellid leeches. A number of radial muscle fibres more or less fan-shaped and 15 to 20 in one plane (Text-fig. 5, P.) stretch from the periphery of the proboscis to the epithelium lining the lumen. They frequently surround the longitudinal muscle fibres at their expanded ends. Midway between the periphery and the lumen of the proboscis are a series of annular muscle fibres. The epithelium lining the lumen of the proboscis is well developed. There is little difference in the muscular structure of the proboscis from that figured by Sukatschoff for Branchellion and by Johansson (1896) for Callobdella lophii and Abranchus brunneus. In the spaces between the radial muscles are placed the ducts of the salivary glands and the blood-vessels of the proboscis.

When the proboscis is retracted it lies surrounded by its sheath in the anterior lacuna, its apex lying close to the subcesophageal ganglion.

The lumen of the proboscis opens posteriorly into a slight expansion of the entodermal anterior gut, which has been called the bulb in Branchellion.

The gut narrows immediately and gives off in somite 11 apair of œsophageal glands (Text-fig. 6, *Œs. gl.*). These lie in somites 10, 11, and 12, and communicate with the œsophagus by a very narrow lumen (5 to 10  $\mu$  in diameter) in the eleventh somite just above the nerve ganglion. These glands are placed dorso-lateral to the accessory male glands and are somewhat convoluted. They measure '3 mm. in a longitudinal direction, and may, when distended, measure '1 mm. transversely. Glands of this nature were first described by Johansson (1896) for the Ichthyobdellid leeches Piscicola, Callobdella, and Abranchus.

In 1913 Sukatschoff described similar glands in Branchellion, and compares them to the œsophageal glands of Hæmenteria costata described by Kowalevsky (1900), and to similar glands of Clepsine plana figured by Whitman (1891); Hemmingway (1912) found them also in the Glossiphonid leeches Placobdella pediculata and P. parasitica.

Both Johansson and Sukatschoff state that they are glandular organs and that fish blood-corpuscles are not found in them, and I agree with them.

The nuclei of the epithelial cells which line these glands are of the same character as those of the following pouches of the thin-walled parts of the middle gut, but they are larger, some being twice the size.

At the level of the opening of the female pore that is just below the 12th nerve ganglion the second pair of pockets of the anterior entodermal gut is given off (Text-fig. 6, A.g.). They have wide openings into the gut and are somewhat convoluted at their blind ends. These pouches appear to correspond to those figured in Branchellion by Sukatschoff (fig. 85, V. d. t. 2), and are homologous with the following six pairs of the anterior thin-walled division of the middle gut. Their difference in form is caused by their being pressed forward by the cocoon gland ducts, which prevent lateral distention. Behind this pair of pouches there is a sphincter of annular muscle fibres.

Following the anterior entodermal gut is the middle gut, which is divided into-

- (1) An anterior thin-walled part (Text-fig. 6, A.m.g.).
- (2) The blind gut (Text-fig. 6, B.g.).
- (3) A dorsal thick-walled part (Text-fig. 6, M. g.).

The anterior thin-walled division of the middle gut lies in the testicular region, and consists of a small median tube from which are given off six pairs of cæca opposite to nerve ganglia 14 to 19 inclusive. The position and form of these six pairs of cæca are shown in Text-fig. 6, A.m.g.

These pouches are lobed at their blind ends by the presence

of dorso-ventral muscle fibres, which produce two or more lobes in a distended pouch. A sphincter of annular muscle fibres surrounds the tube just before and immediately behind the first pair of pouches, and dorso-ventral muscle fibres are also present about the neck of the following pouches and may act as sphincters. A similar arrangement has been described by Sukatschoff in Branchellion, but Johansson (1896) states that all the pouches of this part of the gut of Callobdella nodulifera have sphincters of annular muscle fibres.

The anterior thin-walled part of the middle gut is followed ventrally by the blind gut (Text-fig. 6, B.q.), which in Austrobdella consists of a pair of elongated pouches which open into the last pair of cæca of the thin-walled part of the middle gut. They extend back to the anal region and are fused with one another in five places. In fact the fusion is so complete that the parts which are not fused form merely four small apertures. When this blind gut region is filled with fish corpuscles there is seen on either side a series of cæca extending laterally, which decrease in size from before backwards. The extent of the development of this blind gut has an important bearing on an interesting hypothesis put forth by Johansson (1898). All the thin-walled parts of the middle gut are lined by a single layer of flat epithelial cells, which become considerably stretched when a pouch is distended, but are well developed in the median parts. Weak muscle fibres are occasionally present, and the pouches are generally filled with undigested fish blood-corpuscles.

A comparison of the blind gut region of Austrobdella translucens with Johansson's (1898) figures shows that here the development of the blind gut is intermediate to that found in Callobdella nodulifera and C. lophii. In this paper Johansson compares the structure of this blind gut region in the different Ichthyobdellids and divides them into three types. The first, and in his opinion the most primitive, type is represented exclusively by the genus Abranchus, which has two completely, or almost completely, separated blind pouches. The second type, represented exclusively by the

genus Pontobdella, has a single large undivided blind pouch. The third type, to which all remaining genera belong, shows transition forms between these two types. They have blind pouches fused for a greater or less extent in five places. In this group he placed the genera Platybdella, Piscicola, Cystobranchus, and Callobdella, and by the researches of Sukatschoff (1912) Branchellion must be included along now with Austrobdella. The development of the structure of the blind gut seen in the second and third types Johansson correlated with the fact that the leeches possessing it are enabled to exist for some time away from a host by reason of the greater storage capacity produced by this partial or complete fusion: and with the fact that it is found along with well-develoded musculature indicating good swimming power in such genera as are likely to experience difficulty in finding a fresh host. To this likely hypothesis as to the cause of the development of this fusion of the blind pouches Austrob della affords little support. In fact, the case presented here is almost as hard to fit in with Johansson's hypothesis as that of Callobdella lophii, to explain which Johansson (1898) has to say that "it is hardly too bold to think that this leech never leaves its host." He offers no explanation as to how the cocoons are deposited, though elsewhere he states that all Ichthyobdellids, so far as is known, deposit their cocoons away from their hosts. Here I may mention a note by Leigh-Sharpe (1913) about the capture of a large angler (Lophius piscatorius), only a few hundred yards from shore, with five specimens of C. lophii. Now in Austrobdella the blind gut is better developed than in C. lophii; the musculature is weak (about the same as C. lophii), and the ability to swim absent. The sand whiting, on which Austrobdella is exclusively parasitic, so far as I can ascertain, is common, and lives in very shallow water and feeds along shoals and beaches, frequently burrowing in the sand. According to this mode of life, following the reason used by Johansson for Abranchus, there should be little need for extra storage of food, yet in Austrobdella I find a development of the blind gut about

half that of Callobdella nodulifera, a species parasitic on a deep-water fish; however, the latter has better muscular development. It seems that the explanation of these apparent exceptions to what appears to be a well-reasoned hypothesis can only be gained when the life history of these leeches is discovered. The only record of Austrobdella translucens being found away from its host is a curious one, a specimen being found by Prof. J. P. Hill some years ago in the gastric pouch of a jelly fish (Cambessa mosaica).

The dorsal thick-walled part of the middle gut .--In Austrobdella only three pairs of pouches are developed in this division (Pl. 1, fig. 1, M. g.). The fourth pair of thickwalled pouches is present only in a rudimentary state. The first pair of pouches lies between the 19th and 20th nerve ganglia (Text-fig. 6, M.g.); the fourth, which in other Ichthyobdellid leeches is placed between the 22nd and 23rd nerve ganglia, is represented here by a flexure of the intestine (Text-fig. 6, M, q, f.) The cells of the epithelium lining this part of the gut have their free surfaces covered with a film of densely-placed cilia, so that they present a fringed appearance in section, similar to that described by Sukatschoff as occurring in Branchellion (1912, Fig. 78). There does not appear to be present a section of the gut bearing a ciliated epithelium of the usual type, such as occurs in Branchellion and in other Ichthyobdellids (Johansson, 1896 a). Following the thick-walled part of the middle-gut there is the posterior gut, formed chiefly by the rectum (Text-fig. 6, R.), which opens by the anus on the dorsal surface of the 27th somite. The walls of this portion of the gut are muscular and the epithelium is similar to that lining the thick-walled part of the middle gut, but is not ciliated.

#### THE BLOOD-VASCULAR SYSTEM.

Following the well-known work by Oka in 1894, dealing with the blood-vascular system in Clepsine, there appeared the investigations of Johansson (1896 b) and Selensky (1906).

relating to this system in Piscicola. In 1902 Oka published a paper, in which he summarised his investigations concerning the blood-vascular system in the various families of the Hirudinea. In a lucid manner he showed that only in the Glossiphonidæ and Ichthyobdellidæ was a true bloodvascular system present and that it had no communication with the lacuna system. Again, in 1904, Oka described in some detail the vascular system in Ozobranchus. My investigations of Austrobdella have shown that a closed blood-vascular system is also present here.

In general this system in Austrobdella resembles that described in Ozobranchus and differs from the Piscicola and Callobdella type.

There are, however, several important differences from Ozobranchus. The lateral paired branches in the anterior part of the body are three as compared with the four pairs found in Clepsine and the Ichthyobdellid leeches so far described. It is the second pair which are wanting. There is also a ring vessel in the posterior sucker with which the loops from the dorsal and ventral vessels connect; this is a very different arrangement from any so far described.

Lastly, the division of the dorsal vessel into two parts takes place in the 24th somite, which is much higher up than in Clepsine and Piscicola.

I have been favoured in these observations on the bloodvascular system by the transparent nature of the leech. The diagram of the blood-vascular system (Pl. 1, fig. 2), is a careful representation of the course and relations of the blood-vessels in the neck and anterior sucker.

The course of the blood-vessels is as follows :

The dorsal blood-vessel gives off, in the anterior part of the body, three pairs of lateral branches, and an unpaired proboscis branch. The first of these (Pl. 1, fig. 2, L.v. 1.), is formed by the forking of the dorsal vessel in the oral sucker. The two branches given off run round the eye-spots and unite in the region of the subœsophageal ganglion to form the ventral blood-vessel. The course of this first pair of lateral

vessels is very similar to that found in Ozobranchus (Oka, 1904, Fig. 1). The second pair of lateral vessels (L. v. 2), is given off in somite 9; these branches run ventrally and anteriorly, and unite with the ventral vessel just behind the spot where the first pair join. The third pair of lateral vessels (L. v. 3) are given off in somite 10; the two branches run at first posteriorly to the end of somite 11; then they curve sharply and, running forward, enter the ventral vessel, just behind the point of entry of the second pair of lateral vessels.

Immediately in front of the first valve the dorsal vessel gives off the proboscis branch (P. v. 1.) which runs to the apex of the proboscis and there bifurcates. The two vessels thus formed unite almost at once to form the efferent proboscis branch (P. v. 2), which runs to join the ventral vessel, just behind the point where the second pair of lateral vessels enter into it. After emerging from its intimate relations with the cæca of the thick-walled intestine posteriorly (I. s.), the dorsal vessel divides in two at the beginning of the 24th somite. These branches extend in such a way as to form a vessel running round the periphery of the posterior sucker. This part of the dorsal vessel gives off on each side four or five short-looped vessels, which communicate with a ring vessel (R.v.) running right round the periphery of the posterior sucker. This ring vessel receives on each side seven branches of the ventral vessel. A certain degree of anastomosis is seen in these branches before they unite to form the ventral vessel. I have followed the course of this ring vessel in living leeches obtained from different places.

As stated in the account of the cœlomic system, the dorsal blood-vessel lies in the dorsal lacuna, or its extension the intestinal lacuna, for almost the whole extent of these lacunæ. Anteriorly the dorsal vessel passes out of the lacuna at somite 13. Pl. 2, fig. 12, shows the vessels in the 11th somite shortly after the dorsal blood-vessel has left the lacuna. The ventral vessel is lying free in the dilated lacuna which surrounds the accessory male glands.

Posteriorly the two branches of the dorsal blood-vessel in the 26th somite leave the lacuna, formed by the union of the intestinal and ventral lacunæ, and enter the connective tissueto course round the periphery of the posterior sucker.

The ventral vessel lies in the ventral lacuna from the 7th to the 26th somite. It is formed by the union of the first pair of lateral vessels, just above the 7th somite. These lateral vessels enter the lacuna opposite the spot where they fuse. The ventral vessel lies quite free in the ventral lacuna above or at the side of the nerve-cord and always above the nerve ganglia. It leaves the ventral lacuna near the same spot as the dorsal vessel in the 26th somite.

The histological features of the blood-vessel walls agree closely with those described for Callobdella by Johansson (1896 b) and for Piscicola by Salensky (1906).

The dorsal vessel, immediately on passing out of the dorsal lacuna, develops in the 13th somite a strong muscle layer of annular nature internal to a layer of finer muscular fibres. This structure continues until the diameter of the dorsal vessel becomes smaller after the proboscis branch has been given off. The anterior part of the dorsal vessel takes its origin in a peculiar way from that part of the dorsal vessel which has a much greater diameter. On the side of the dorsal vessel, opposite to the point of origin of the proboscis vessel, this anterior part lies laterally to the dorsal vessel and opens into it at two places, both of which are guarded by valves. The lateral vessels, possessing this well-developed muscle layer for but a little distance after they spring from the dorsal vessel, gradually come to resemble the ventral vessel in the nature of their walls. The wall of the dorsal bloodvessel in the dorsal lacuna possesses only a thin epithelium, with scattered nuclei (Pl. 2, fig. 10, Nu. epi.), save only at those places where the valves are placed. Here there are one or two annular muscle-fibres, such as are found in the preclitellar region, and which, in contraction, form a sphincter, against which the valve is pressed by the backward pressure of the fluid during the contraction of the dorsal vessel (Pl. 2,

fig. 11, S. m.f). In somite 19 the dorsal vessel enters into intimate connection with the cæca of the thick-walled intestine. This remarkable arrangement was first described by Johansson for the Ichthyobdellids in Callobdella, and in many respects the relations of the blood-vessel with the gut-walls, as described by him, hold good also for Austrobdella.

Text-fig. 3 shows the relations of this vessel with the epithelium and muscular walls of the gut. At the beginning of the thick-walled intestine, the dorsal vessel is seen connected with the muscular layer of the gut, and almost immediately it opens on either side into the intestinal sinus, and ceases to be distinguishable from the walls of the sinus.

The intestinal sinus is formed by the separation of the epithelial and muscular walls of the gut. This separation is not complete in Austrobdella, for here and there the normal relations of the epithelial layer and the muscle layer are seen (Text-fig. 3, M. g. ep.), but save at these places of attachment, which are usually small, the blood-stream surrounds the epithelial layer of the thick-walled intestine. These relations are such as described for Callobdella. In the intermediate portions of the thick-walled intestine, which connect the paired pouches, the dorsal blood-vessel separates from the intestinal sinus and lies in the dorsal side of these regions. Also, in the region of the fourth rudimentary pair of cæca, the sinus developed from the dorsal vessel is very small, and only for a short distance does the dorsal vessel cease to be defined: following this part the dorsal vessel is clearly defined and remains single until above the ganglion of the 24th somite; here it divides in two. The two branches then run laterally to the gut closely connected with its muscular wall. In the 27th somite these two vessels diverge and run round the periphery of the posterior sucker on either side and finally unite (Pl. 1, fig. 2). The valves in the dorsal vessel are found from just before the giving off of the second pair of lateral vessels to the beginning of the intestinal sinus (Pl. 1, fig. 2, Vl.). They are placed somewhat irregularly, one or two in each somite. They are generally shaped

like a fir cone, measuring 80  $\mu$  long, and are made up of about eight cells. In some cases these cells become separated and present the appearance in section shown in Pl. 2, fig. 11. The valve placed just before the intestinal sinus is twice the length of the others.

## Circulation of the Blood.

I have investigated the circulation of the blood and the lymph in living specimens found on whiting which I caught on the ocean beach. The chief mechanism for propelling the blood is the peristaltic contraction of the muscular wall of the intestinal sinus, first described by Johansson (1896). This peristalsis occurs in the three pairs of pouches of the thick-walled intestine and also in the rudimentary pouch. In the living animal the whole of the thick-walled intestine is in a state of active contractile movements. These begin where the thick-walled intestine passes into the rectum. The general movement is from behind forwards. The whole of the contractile muscular wall appears to contract simultaneously when the animal is very active, but in specimens in which the rate of contraction is lowered it is seen that the peristalsis is There is seen, however, a certain from behind forward. amount of individual contraction of separate paired cæca. The most active specimens I examined showed a rate of contraction of over thirty times per minute. The blood forced forward by the contractions is prevented from flowing back by the valves of the dorsal vessel.

In the dorsal vessel the backward pressure of the blood causes the valve to press against the sphincter fibres of the dorsal vessel just behind it. Very rapidly the blood passes onward, and the next valve acting in the same manner, the first valve is again forced forward by the incoming blood. The constriction of the dorsal vessel is much greater at the sphincter muscle fibres than elsewhere. So great indeed is the contraction here that the dorsal vessel resembles a string of sausages. The constrictions being at the points occupied

by the sphincters (Pl. 1, fig. 2, Vl.), the dorsal vessel in the testicular region is caused to contract by the pressure of lymph in the lacuna, so that its walls come into contact. In this manner the blood is forced forward and ultimately enters the paired branches; these are not provided with valves. The blood then passes along the non-contractile lateral vessels into the non-contractile ventral vessel and so through the complicated branches in the posterior sucker, until it again reaches the intestinal sinus. The important relation of the contractions of the muscular walls of the intestinal sinus to the flow of the lymph and the contractions of the contractile lacuna has been dealt with in the section describing the circulation of the lymph.

## NEPHRIDIAL SYSTEM.

There are eleven pairs of nephridia arranged segmentally in the 13th to the 23rd somite. These form what is practically a continuous network in this area. However, this is so arranged that the segmental character of the nephridia is obvious. An inspection of Pl. 2, fig. 7, will make this clear. The best developed parts of the nephridia are two tubes (L. n. c.), which are placed ventral to that part of the lacuna formed by the fusion of the dorsal and ventral segmental lacunæ. These tubes have a diameter of about 40  $\mu$ , while the diameter of their lumen is 5  $\mu$ . They pursue a tortuous course and frequently branch, and the branches anastomose. They give off in each somite branches which run to open at the nephridiopore (Np.), and they receive the branches which run from the dorsal and ventral networks.

The paired branches, which are given off in each somite to open at the nephridiopore, are of the same size as the chief branches of the lateral canals. They are given off from the lateral canals near the level of the first annulus of the segment, and run medially and posteriorly, curving sharply as they approach the ventral lacuna at an angle of  $45^{\circ}$  (Np. b.). They run laterally for about half the distance of their first course

and open on the second annulus of each segment. The diameter of the aperture of the nephridiopore is about 5  $\mu$ . In each segment the lateral canals receive three or four main branches, which are the outcome of the anastomotic canals. These latter are best developed around the ventral lacuna, but they also surround the dorsal lacuna. The arrangement and relative size of these canals is shown in Pl. 2, fig. 7.

As is general in Hirudinea, the canals of this nephridial system are intracellular. The cells, which are burrowed through by these canals, have oval nuclei.

The nephridiopores open directly on the body surface and not into pits, like those of Cystobranchus.

Neither in the living animal nor in serial sections have I been able to find internal openings, so that the ciliated funnels, possessed by Branchellion and Pontobdella, are here absent. In this respect then Austrobdella resembles Piscicola, Callobdella, Cystobranchus, Abranchus, and possibly Platybdella.

This nephridial system is most like that described by Johansson for Callobdella. It differs in the much greater development of the branch going to the nephridiopore, and the greater degree of anastomosis of the smaller channels.

In Austrobdella there exists on either side a fine canal, which may represent the dorso-lateral canal described for Callobdella. The lateral tubes are well developed in Austrobdella, being  $40 \,\mu$  as compared with Abranchus and Callobdella  $20 \,\mu$ , Piscicola  $30 \,\mu$ , but again are smaller than Cystobranchus  $50 \,\mu$  (Johansson, 1896).

#### REPRODUCTIVE SYSTEM.

Thanks to the excellent work of Brumpt (1900), 'Reproduction des Hirudinées,' it is possible to compare the reproductive organs of Austrobdella with those of allied leeches. This species in its male organs resembles most Callobdella (Trachelobdella) lophii as regards the structure of the ejaculatory canal, but it lacks the muscular organ of Johansson

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and the conducting tissue of the bursa described by Brumpt. Of the spermatophore glands, those described by Brumpt as A and B glands are present, while I am doubtful as to the presence of C glands. The glands A are well developed, and are enclosed in the muscular tunic of the terminal parts of the ejaculatory canals. The glands B, and perhaps C, surround the terminal parts of the ejaculatory canals and open into the common part. Here the resemblance, owing to the development of the A glands, is more to the Glossiphonid type than to C. lophii.

In the female organs, owing to the isolated ovaries and the absence of both copulatory area and conducting tissue, Austrobdella resembles Callobdella lubrica, Platybdella soleæ, and Glossiphonia complanata.

Concerning the interesting fertilisation by means of hypodermic injection of spermatophores, which Brumpt has shown to be true in most marine leeches, I have not yet ascertained if a similar phenomenon occurs in Austrobdella. I hope later again to keep these leeches in captivity and to endeavour to bring about copulation with a view to determining this point.

There are five pairs of testes (Pl. 1, fig. 1, T., Text-fig. 6, T.) placed in somites 14-18 inclusive. The vasa deferentia (Pl. 2, fig. 12A, V. def.) on leaving the connective-tissue, become more than doubled in their diameter and constitute the ejaculatory canals, which lie in the expanded anterior end of the ventral lacuna. These ejaculatory canals become considerably coiled at the level of the 12th nerve ganglion (Pl. 2, fig. 12A, Ei. c.), and the lumen of each increases slightly and forms a seminal vesicle. They then come into close relation with the dorsal blood-vessel (Pl. 2, fig. 12,  $E_{j.c.}$ ) and shortly open dorso-laterally into the terminal parts of either side (Pl. 2, fig. 12A, Ej. t.). The terminal parts of the ejaculatory canals are provided with a muscular tunic (Pl. 2, fig. 12, Ej. t.) which encloses the unicellular glands, called the A glands by Brumpt (1900). The two terminal parts open into a common part, the spermatophore sac of

Kovalevsky (1900) (Pl. 2, fig. 12A, Sp. s.), into which open the ducts of the unicellular spermatophore glands called the B glands by Brumpt (Pl. 2, fig. 12, Sp. gl.). The spermatophore sac leads into the bursa-an ectodermal invagination, whose external opening is the male orifice (Pl. 2, fig. 6). The muscular tunic of the terminal parts consists of a single layer of fibres imbricated at their ends. About four to six fibres complete the circuit. At the anterior end the wall becomes two or even three fibres thick. This circular musculature is continued on around the spermatophore sac. but here the fibres, placed two deep, are separated from those on either side by the ducts of the spermatophore glands. At the bursa the fibres decrease to a single layer, and placed external to them are several longitudinal fibres, apparently the diverted fibres of the body-wall muscles round the male genital opening, which may aid in the protrusion of the bursa (Pl. 2, fig. 6). The terminal parts of the ejaculatory canals are lined throughout with gland cells, the ducts of which run radially and are directed towards the spermatophore sac (Pl. 2, fig. 12, Ej. t.). These ducts almost obliterate the lumen into which they pour their secretion. There are found among these ducts the small cells of the supporting tissue.

The cytoplasm of these glands stains more deeply with hæmatoxylin than those of the accessory male glands outside the muscular tunic.

These latter cells make up a well-developed glandular mass, which lies in somites 11 and 12 (Text-fig. 6, Sp. gl.). Each is a unicellular gland (Pl. 2, fig. 12, Sp. gl.) and opens in o the spermatophore sac. On either side this mass presents two lobes caused by dorso-ventrally placed muscle fibres.

The development of these glands is such that they wrap round the terminal parts of the ejaculatory canals (Pl. 2, fig. 12, Sp. gl.).

The secretion space of each cell is generally filled with numerous granular eosin-staining particles, as are also the ducts opening into the spermatophore sac.

I have not determined, by the staining methods of Brumpt, if there are two kinds of gland cells present; but in sections stained by hæmatoxylin and eosin they appear to be all of one kind.

The bursa, which is shown in Pl. 2, fig. 6 (a medial sagittal section) is lined by a continuation of the epidermis of the body.

The ovaries, which are two simple sacs lying free in the ventral lacuna, show considerable movement in the living animal-a fact which Moquin-Tandon (1846) says caused Rondeau to take them for hearts in certain leeches. They become united just above the 13th nerve ganglion, and from their junction the oviduct (Pl. 2, figs. 6, 12A, Ovd.) runs dorsally and curves to enter the glandular part of the bursa (Pl. 2, figs. 6, 12A, B. gl.). After their junction, the ovaries are continued forward to form each an anterior horn. The oviduct, which has a small lumen (Pl. 2, figs. 6, 12A, Ovd.). has a wall of circular muscle fibres, and external to these a well-developed connective tissue layer with large nuclei, such as Brumpt has described as general in Ichthyobdellids, and through which, he says, spermatozoa are frequently seen working their way.

The epithelium lining the vagina is a continuation of the epidermis of the body, which is thrown into folds in the glandular part of the bursa.

The ovaries in adult specimens are filled with ova in various stages of development. The development of the ovam (Pl. 2, fig. 6, Ov.) and the breaking-down of the yolk cells appear to be similar to what occurs in Callobdella lophii, as described by Brumpt. Near the oviduct the ova are seen undergoing the first maturation division.

This work was begun while holding the John Coutts' Scholarship. I wish to thank Prof. W. A. Haswell, in whose laboratories I worked, for his advice and kindness. Mr. F. W. Atkins, of the Sydney Technical College, re-drew the figures, and Mr. W. Graham, of the Department of Zoology here, gave me great assistance with photographs of the living specimens.

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## EXPLANATION OF PLATES 1 AND 2,

Illustrating Mr. C. Badham's paper "On an Ichthyobdellid parasitic on the Australian Sand Whiting" (Sillago ciliata).

[All figures refer to Austrobdella translucens.]

#### PLATE 1.

Fig. 1. — Drawing from life. Seen from the ventral aspect by transmitted light. The specimen is somewhat extended so that the shouldered appearance seen in large individuals (vide fig. 5) is absent. The pigment cells (P. c.) are represented and the ejaculatory canals (Ej. c.) show up on account of their pigmentation. The five pairs of testes (T.) are a marked feature. Certain parts of the Blood-vascular System appear, and the thick-walled part of the middle gut stands out clearly (M. g.). A. m. g. Cæca of anterior thin-walled part of middle gut. C.1. Contractile lacuna. Cl. gl. Clitellar glands. E. Eye spots. Ej. c. Ejaculatory canal. M. g. Cæca of thick-walled part of middle gut. Ov. Ovary. P. Proboscis. P. c. Pigment cell. T. Testis. V. v. Ventral vessel.

Fig. 2.-Drawing of the Blood-vascular System from life. The dorsal vessel is shaded, the ventral vessel is outlined. The origin of the dorsal vessel is seen in the posterior sucker; just above the union of the two branches from the posterior sucker this vessel enters into intimate relation with the cæca of the thick-walled intestine to form the intestinal sinus (I. s.). After leaving the sinus the dorsal vessel (D. v.) has a series of values (Vl.), one being placed where a constriction is seen on contraction. Anteriorly, the dorsal vessel gives off two pairs of lateral vessels (L. v. 3, L. v. 2) and an impaired proboscis branch (P. v. 1), and bifurcates just above the level of the eye spots to form the first pair of lateral vessels. The paired lateral branches are gathered together to form the ventral vessel (V, v). This runs posteriorly and gives off in the posterior sucker a series of anastomotic branches which go to form the ring vessel (R. v.). D. v. Dorsal vessel. E. Eye spots. L. v. Lateral vessel. P. v. 1. Afferent proboscis vessel. P. v. 2. Efferent proboscis vessel. R. v. Ring vessel. Vl. Valve.

Figs. 3, 4, 5.—Drawings of alcoholic specimens (fixed in boiling corrosive acetic), 1.5 mm., 4 mm. and 9 mm. long respectively. Fig. 3

is a side view, the other two figures are ventral views. This series shows the change from the cylindrical form of the young specimen (fig. 3) to the slightly flattened sexually mature specimen 4 mm. long (fig. 4). The shouldered appearance of the body of the specimen 9 mm. long (fig. 5) and its greatly flattened form are in striking contrast with the form of the younger specimens.

#### PLATE 2.

Fig. 6.—A medial sagittal section of a specimen 8 mm. long, showing the genital openings, bursæ, oviduct, ovaries, spermatophore sac, and the relations of the ventral nerve cord and the ventral and dorsal blood-vessels in this region. Compare with fig. 12A. The male genital opening is seen at 3 and the bursa leading into the spermatophore sac (Sp. s.). The female genital opening is seen at  $\mathcal{Q}$ leading into the bursa, which has the walls of its dorsal part plicated (B, gl.) to form the glandular part of the female bursa. The oviduct (Ovd.) has been cut at two places, where it leaves the junction of the ovaries and more dorsally where it is about to enter into the glandular part of the bursa. The sections of the ovaries (Ov.) show ova in various stages of formation by the breaking down of yolk cells; the two spindles represent maturation divisions. The relations of the ventral nerve cord and ganglia 11, 12, 13 to the sexual organs are shown, and may be compared with the model depicted in fig. 12A. The ventral vessel (V. v.) is seen lying free in the expansion of the ventral lacuna which contains the sexual organs in this region. The dorsal vessel (D. v.) is seen as it is entering the dorsal lacuna. The degree of development of the circular and longitudinal body-wall musculature is displayed. B. gl. Glandular part of female bursa. C. m. Circular body-wall musculature. D. v. Dorsal vessel. L, m. Longitudinal body-wall musculature. N. gn. Nerve ganglion. Ovd. Oviduct. Ov. Ovary. Sp. s. Spermatophore sac. V. v. Ventral vessel.

Fig. 7.—Diagram in relief of the Nephridial System in the testicular region of the body viewed from the ventral surface. The posterior end of the leech would be towards the observer. For the sake of clearness the somite is shown as consisting of the three primitive annuli. In order to show the segmental nature of the nepridial system one whole somite and its portion of the nephridial system is drawn, and parts of the preceding and succeeding somities with their portions of this system. The ventral and dorsal lacuna are shown, but neither the segmental nor the contractile lucunæ. On either side the lateral nephridial canal is seen receiving branches of the fine capillary network of nephridial tubules which extend in the dorsal and ventral sides of the body. The lateral nephridial canals are shown giving off in each

somite the pair of branches (Np. b.) leading to the nephridiopores (Np.). D. l. Dorsal lacuna. D. v. Dorsal vessel. L. n. c. Lateral nephridial canal. N. p. Nephridiopore. Np. b. Duct leading to nephridiopore. T. Testis. V. l. Ventral lacuna. V. ne. Ventral nerve cord. V. v. Ventral vessel.

Fig. 8.—Diagram in relief of the Lacuna System, viewed from the dorsal surface, in a somite of the testicular region of the body. The dorsal lacuna (D.l.) is shown containing the dorsal vessel (D.v.). In the ventral lacuna (V.l.) are the ventral nerve cord (V.ne.) and ventral vessel (V.v.). The dorsal and ventral parts of the segmental lacuna of either side of the somite are seen to junction laterally, and after bifurcating to join up with branches from the preceding and succeeding segmental lacuna of the same side. The segmental lacuna opens into the contractile lacuna in two places in each somite. The contractile lacuna receives in each somite three capillaries dorsally on either side. Cap. l. Capillaries opening into contractile lacuna. C.l. Contractile lacuna. D. l. Dorsal lacuna. D. v. Dorsal blood-vessel. S. l. Segmental lacuna. V. l. Ventral lacuna. V. v. Ventral vessel.

Fig. 9.—Drawing of horizontal section through the testicular region of the body showing the character of the contractile lucana (C.l.) and the branches of the segmental lacuna (S.l.) leading to it. In one place a branch opens into the contractile lacuna and the opening is guarded by sphincter muscle fibres (S. m. f.). The large unicellular lateral glands (L. gl.) are shown, and the clitellar glands medial to the contractile lacuna. C.l. Contractile lacuna. Cl. gl. Clitellar glands. L. gl. Unicellular lateral glands. (S. m. f.) Sphincter muscle fibres. S.l. Segmental lacuna.

Fig. 10.—Drawing of a transverse section through the dorsal bloodvessel and dorsal lacuna in the testicular region of the body, showing the dorsal and ventral septa (Sep. d., Sep. v.). The nuclei of the epithelial cells of the dorsal vessel bulge into the dorsal lacuna. D.v.Dorsal vessel. D.l. Dorsal lacuna. Epi. nu. Nucleus of epithelial cell. Sep. d. Dorsal septum. Sep. v. Ventral septum.

Fig. 11.—Drawing of a section through a valve of the dorsal bloodvessel in the testicular region of the body. Usually the valve (Vl.)is fir-cone shaped and when forced back rests against the sphincter muscle fibre (S. m. f.), but sometimes, as shown here, the valve becomes broken up into separate cells attached to a common stalk. On contraction of the dorsal vessel several cells of the valve may be forced past the spincter, as is shown here. D.v. Dorsal vessel. Vl. Valve. - S. m. f. Sphincter muscle fibre.

Fig. 12.—Drawing of a transverse section through the neck region

of a specimen 7 mm. long. The section passed through the eleventh nerve ganglion and is cut obliquely so that the terminal part of the ejaculatory canal (Ej.t.) is shown on the right side and the main mass of the spermatophore glands on the left (Sp.gl.). The terminal parts of the ejaculatory canals are seen just before they have united to form the spermatophore sac (compare with fig. 12A). The ventral vessel is seen lying free in the ventral lacuna, here expanded to contain the sexual organs. The relations of the ejaculatory canals, œsophagus and dorsal vessel in this region are also shown. D.v. Dorsal vessel. Ej.t. Terminal part of ejaculatory canal. Ej.c. Ejaculatory canal. N.gn. 11. Nerve ganglion 11. Es. Esophagus. Sp.gl. Spermatophore glands. V.v. Ventral vessel.

Fig. 12A.—Diagram in relief of the Reproductive System. The terminal part of the right ejaculatory canal (Ej. t.) is shown as cut away for the sake of clearness. The relations of the ventral nerve cord and ganglia to the male and female organs are made evident. B. gl. Glandular part of the female bursa. Ej. c. Ejaculatory canal. Ej. t. Terminal part of the ejaculatory canal. Ovd. Oviduct. Ov. Ovaries. Sp. s. Spermatophore sac. V. def. Vasa deferens. V. ne. Ventral nerve cord. 11, 12, 13. Nerve ganglia. J Male opening.  $\Im$  Female opening.